ANAPLASMA MARGINALE.

DR. A. THEILER.

In the Annual Report of the United States Department of Agriculture for the years 1891 and 1892, Smith and Kilborn published their investigations into the cause of Texas fever, which was found to be due to the presence of an endoglobular parasite and to which they gave, at that time, the name of *Pirosoma bigeminum*. They described two forms of the disease : the acute and the mild one, which latter they also called the autumnal form. The differentiation of these two diseases was based on the aspect the parasites took in the red corpuscles which, although differing in shape and size, were considered to belong to two phases in the cycle of development. Accordingly they distinguished the pear-shaped parasite now called *Piroplasma bigeminum* and which they identified with the acute form of Texas fever, from the second form, the peripheral coccuslike body of the mild or autumnal form of Texas fever. Smith and Kilborn based on these observations the possible life cycle of *P. bigeminum* of which they described three stages :—

- 1st. The (hypothetical) swarming stage, the form of which, however, as they state, could not be traced.
- 2nd. The stage of the peripheral coccus-like bodies, which bodies they thought would develop into the

3rd. The spindle or pear-shaped stage.

Smith and Kilborn already noticed that their third stage is the one which is usually met with in acute Texas fever. They had, therefore, to explain the absence of the coccus-like bodies in the acute stage, and their explanation was that the presence of the coccus-like bodies may be so ephemeral that they escape observation. Under the influence of the temperature of the autumn the second stage would remain as such and not develop into the third one. In this second stage they would cause the mild disease.

To understand this explanation it may be stated here that Smith and Kilborn undertook their experiments usually in July, August, and September, and it was during September, October, and November, that they met with the disease due to the coccus-like bodies. In subsequent observations made in South America by Knuth, the same coccus-like bodies were seen again. This author did not support the view of Smith and Kilborn about the three different stages, yet, nevertheless, he considered them to belong to the life cycle of *P. bigeminum*. On the other hand, Djunkowsky and Luhs, who were studying the piroplasms in the Transcauscus, came across the same parasite; they had noticed the presence of *P. bigeminum* in that country, but they did not identify the coccus-like body with this disease, but with another one, which they called tropical piroplasmosis, and which is due to a small piroplasm called *P. anulatum* very likely identical with our *P. parvum*.

In South Africa I have seen these parasites during a number of years. The American literature not being available to me at the beginning of my investigations, I described them as marginal points in my various reports. The observations which I made led me to think that these marginal points had nothing to do with *P. bigeminum*, but that they represented an independent parasite genus of their own and that they were the cause of a definite disease, which had to be separated from redwater. I was, however, until recently, unable to give this proof in such a way as would remove all doubts.

Some years ago I sent ticks to England (*Boophilus decoloratus*) which were infected with *P. bigeminum*. They produced the disease in London when placed on an ox, and this ox formed the starting point of many investigations and inocculation experiments undertaken in England. As a result of such investigations, Nuttall described the cycle of development of *P. bigeminum* in the blood, and according to this author, it is a simple division, as in the case of *P. canis*. He does not mention any forms corresponding to the coccus-like or marginal points mentioned before.

In Germany the disease haemoglobinuria of cattle was also investigated during the last few years; it was found to be due to a piroplasm which, owing to some slight difference, is considered to be a species of its own, and is called *P. bovis*, but it is very closely allied to *P. bigeminum*. No parasites resembling the peripheral coccus-like bodies were noticed in the life cycle of this parasite.

Stockman and myself have carried out experiments for a number of years to immunize English cattle. The cattle were inoculated in England against South African redwater with the strain of redwater forwarded by means by the already mentioned ticks; after the inoculation the animals were sent to the Transvaal to be exposed here. In the first lot of cattle exposed, I noticed the appearance of marginal points in the blood of two animals, which succumbed to the disease. A second lot of ten heifers, which were all immunized in the same way in London, were not exposed to natural infection after their arrival, but inoculated a second time in the Transvaal with blood containing P. bigeminum. The result was that this inoculation did not break the immunity against redwater. After a certain period, varying in length from twenty-seven to thirty-two days, a typical fever reaction commenced which was initiated and accompanied by the presence of marginal points. Of ten animals injected, five died. It was evident that P. bigeminum was not responsible for this disease. With the inoculation of the blood two different parasites had been injected—P. bigeminum, against which the animals were immune, and another one, the marginal points, which caused the disease. If this conclusion was right, it had to be expected that it would be possible to separate these two parasites, at least to isolate the one with the shorter incubation time. It was, indeed, possible to do so with P. bigeminum, whose incubation period is the shorter one. A pure infection resulted after injections of blood, taken at the beginning of the acute attack of redwater, into a susceptible beast. When this animal, at a later period, was infected with marginal points it promptly reacted to this infection.

The reverse experiment was not so easy, because wherever we had an infection with marginal points it was complicated with *P. bigeminum*, so that the inoculation of such blood would constantly cause redwater in the first instance. An observation I had made showed that animals imported from Aliwal North, although promptly reacting to redwater, did not do so to the injection of blood containing marginal points. I concluded, therefore, that these animals are immune against this latter parasite. It had to be expected that when blood of such animals was injected into susceptible animals only marginal points would appear. This has been done in several instances, and in every one, after a typical long incubation time, marginal points alone appeared. After recovery, the animals were injected with *P. bigeminum* of a pure infection; the result was that they showed this parasite after the usual period of incubation. Thus the independence of marginal points to the cycle of *P. bigeminum* was demonstrated in the reverse order. Accordingly, no doubt can any longer be left that these peripheral bodies represent a genus of their own which produces a specific disease in cattle.

I have proposed to give the name Anaplasma marginale to this parasite. This anaplasma is transmitted by ticks, and it is a noteworthy fact that the incubation time by tick transmission is much longer than that after inoculation of the animals with blood; in the experiments carried out by meit varied from fifty-five to seventy-five days. Accordingly, we understand the results of experiments of the Americans, which were started in the summer. They exposed cattle to tick infection; they noticed the appearance of *P. bigeminum* first, having a shorter incubation time, and only later in the year, after the long incubation time, they noticed the appearance of *Anaplasma marginale*.

It has already been indicated that blood of an immune animal is infective; such an animal forms the reservoir of the virus. This is a peculiarity of the piroplasma diseases, to which group anaplasmosis also belongs. It may be of interest to state my opinion that anaplasmosis is probably the disease which the farmer has hitherto called gall-sickness. Up to the present time we know of three different parasites in South Africa which are found in the blood of immune cattle—*P. bigeminum*, *P. mutans*, and *Spirochaeta theileri*—and to these will now be added *anaplasma marginale*; they can all be transmitted by the inoculation of blood and by ticks.